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Human Protein Data

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Cover illustration: Molecular model of α_1-proteinase inhibitor (antitrypsin) based on that of intact ovalbumin. In the inhibitor the reactive centre loop (in red) will partially fold back into the A-sheet of the molecule (in blue) hinging on the glutamate at position 342 (first residue in red) that is mutated to a lysine in the common Z mutant. Figure prepared by Dr. C. J. Marshall. (See contribution *Alpha-1-Proteinase Inhibitor* by M. C. Owen and R. W. Carrell.)

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Butyrylcholinesterase

Oksana Lockridge

Synonyms	Serum cholinesterase, plasma cholinesterase, pseudocholinesterase, nonspecific cholinesterase
Abbreviations	BChE, BuChE, CHE, ChE
Classifications	EC 3.1.1.8, acylcholine acylhydrolase
Description	Found in human serum or plasma where it is a soluble glycoprotein synthesized in the liver. Also present in most other tissues including brain, muscle, liver, lining of the blood capillaries, intestinal mucosa. Human red blood cells contain membrane-bound acetylcholinesterase (AChE), an enzyme with very similar properties to BChE. Embryonic tissues are rich in BChE. Belongs to family of serine esterases which have an active site serine and are irreversibly inhibited by organophosphate esters. Very fast hydrolysis rates.
Structure	The enzyme in human serum is a tetramer of four identical subunits arranged as a dimer of dimers. The interchain disulfide bond (Cys 571) is important for stability but unimportant for tetrameric structure as this bond can be selectively reduced and alkylated without changing the molecular weight. Noncovalent, hydrophic interactions hold the four subunits together. Membrane-bound forms are found in muscle, intestinal mucosa, capillaries, brain, but the identity of the membrane anchor is not yet known.
Molecular Weight	Molecular weight of BChE in human serum is 340,000 to 348,000 (ultracentrifugation, Sephadex gel chromatography). Subunit weight of the 574 aa (65,092) plus 9 carbohydrate chains (23.9%) is approximately 85,534. The value is approximate because the carbohydrate weight is not exact.
Sedimentation Coeff.	10.7 S
Isoelectric Point	3.99
Extinction Coeff.	18 (280 nm, 1%, 1 cm)
Enzyme Activity	Hydrolyzes choline esters, for example benzoylcholine, butyrylthiocholine, acetylthiocholine, succinyldicholine, as well as noncholine esters for example heroin, aspirin, alpha-naphthylacetate, ortho-nitrophenylbutyrate. Also hydrolyzes the amide, o-nitroacetanilide. Classified as an acylcholine acylhydrolase.
Coenzymes/Cofactors	None
Substrates	Clinically important substrate is the muscle relaxant succinyldicholine, which is hydrolized by people with usual BChE but not hydrolyzed by people with rare genetic variants of BChE. Substrate useful for measuring enzyme activity is benzoylcholine (0.050 mM benzoylcholine, 0.067 M Na/K phosphate buffer pH 7.4, 25 °C; difference in absorbance at 240 nm between substrate and product is 6,700 $M^{-1}cm^{-1}$). Butyrylthiocholine (1.0 mM butyrylthiocholine, 0.3 mM DTNB, 0.1 M Na phosphate pH 8.0, 25 °C; extinction coefficient of product is 13,600 $M^{-1}cm^{-1}$ at 412 nm) and propionylthiocholine are also widely used. Advantage of benzoylcholine is that it gives linear kinetics and is not hydrolyzed by AChE in red blood cells.

Inhibitors	Naturally occurring inhibitors are eserine, from calabar beans, and solanine from potato peel. Synthetic inhibitors include: organophosphate insecticides, chemical warfare nerve agents, eye drops, carbamates used as pesticides, antiasthmatic bronchodilator (bambuterol), drug to treat myasthenia gravis (neostigmine), drug to treat Alzheimer's disease (tacrine) and psychosis (chlorpromazine). Inhibitors that selectively inhibit BChE include iso-OMPA (tetraisopropylpyrophosphoramid), ethopropazine, and bambuterol. Inhibition by eserine at 10^{-5}M (after 30 minutes preincubation) defines an esterase as a cholinesterase. Inhibitors for classifying genetic variants are dibucaine, NaF, and RO2-0683 (the dimethylcarbamate of [2-hydroxy-5-phenylbenzyl]-trimethylammonium). The mechanism of inhibition by organophosphate esters is irreversible alkylation of the active serine (Ser-198). Carbamates also alkylate the active site serine but inhibition is reversible.
Biological Functions	Biological function is unknown. Role in cell proliferation and differentiation is suggested by its highly specific distribution in developing chicken retina and monkey visual pathway.
Physiology/Pathology	Clinically important for diagnosis of poisoning by insecticides of the organophosphate ester and carbamate types. The poisons are toxic because they inhibit AChE at the nerve muscle junction, not because they inhibit BChE. However, serum BChE activity indicates extent of inhibition of AChE at the nerve muscle junction. Decreased concentration of BChE in human serum accompanies severe liver disorders, such as cancer and cirrhosis, reflecting the diminished capacity of hepatocytes to synthesize proteins. When serum BChE activity falls below 0.2 U/ml (normal is 1 U/ml) the patient will experience prolonged apnea after receiving a single dose of succinyldicholine. Complete absence of BChE occurs naturally in 1 out of 100,000 Caucasians, who have the silent genetic variant. No confirmed health abnormalities have been noted in people with silent BChE. A two or three fold elevation of BChE occurs in another rare genetic variant, the Cynthiana variant, and this also has no obvious consequences, other than a resistance to succinyldicholine.
Degradation	Degradation to monomer, dimer and tetramers depleted in the interchain disulfide bond at Cys-571 occurs as a result of partial digestion by protease. Less than 5% of the enzyme in human serum has the smaller sizes. The smaller forms have activity. The tetramer contains 36 carbohydrate chains of the complex type terminating in sialic acid which suggests that clearance of aged BChE ocurs via the galactose receptor in liver.
Genetics/Abnormalities	One out of 3000 Caucasians is homozygous for the atypical variant, in which aspartic acid 70 has been replaced by glycine. Patients with atypical BChE are unable to breathe for as long as 2 hours after a normal dose of succinyldicholine; this dose produces apnea of 3 to 5 min duration in most people. Other genetic variants that respond with prolonged apnea are silent-1 (BCHE*FS117), fluoride-1 (BCHE*243M), fluoride-2 (BCHE*390V), H variant (BCHE*142M) and J variant (BCHE*497V). The K variant (BCHE*539T) has no problems with succinyldicholine. The Cynthiana variant is resistant to succinyldicholine because of elevated BChE activity. There is a single gene for BCHE in man as well as in monkey, cow, sheep, pig, rabbit, dog, rat, mouse, guinea pig and chicken. The human gene is located on the long arm of chromosome 3 at q26.2.
Half-life	11 days in serum
Concentration	Normal concentration in human serum is 0.005 g/L

Isolation Method	Isolation by ion exchange chromatography at pH 4.0 purifies BChE about 800 fold. 2nd step is affinity chromatography on procainamide-Sepharose. Third step is ion exchange at pH 7.0. Purified enzyme is stored in sterile phosphate buffer at neutral pH at 4 °C. The enzyme in serum as well as the most highly purified enzyme are more stable than partially purified preparations. BChE is very unstable after the 1st step because proteases copurify and the protective effect of albumin is lost; the preparation is more stable after affinity column. Specific activity of highly purified human BChE is 200 µmoles/min/mg (0.05 mM benzoylcholine, 0.067 M Na/K phosphate buffer pH 7.4, 25 °C), or 700 µmoles/min/mg (1 mM butyrylthiocholine, 0.1 M Na phosphate pH 8.0, 25 °C). kcat/Km = 50×10^6 $M^{-1}s^{-1}$ for benzoylcholine; 1.5×10^6 $M^{-1}s^{-1}$ for butyrylthiocholine.
Amino Acid Sequence	Mature BChE in serum has 574 aa per subunit. The catalytic triad consists of Ser-198, His-438, Glu-325. The atypical variant has a reduced affinity for all positively charged substrates and inhibitors, and therefore Asp-70 appears to be a component of the anionic substrate binding site.
Disulfides/SH-Groups	Each subunit contains 3 intrachain disulfide bonds at Cys 65–92, 252–263, and 400–519. Two subunits are covalently linked through a disulfide bond at Cys-571. One free but inaccessible sulfhydryl is at Cys-66.
General References	Lockridge, O. Genetic variants of human serum cholinesterase influence metabolism of the muscle relaxant succinylcholine. *Pharmac. Ther* 1990. **47:** 35–60. Whittaker, M *Cholinesterase. Monographs in Human Genetics*, **Vol 11**, Beckman, L. (ed.) Karger, Basel, Switzerland 1986.
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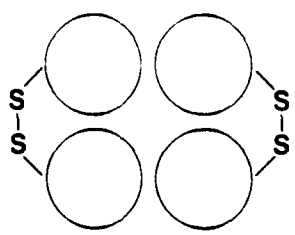

Schematic model of serum cholinesterase.